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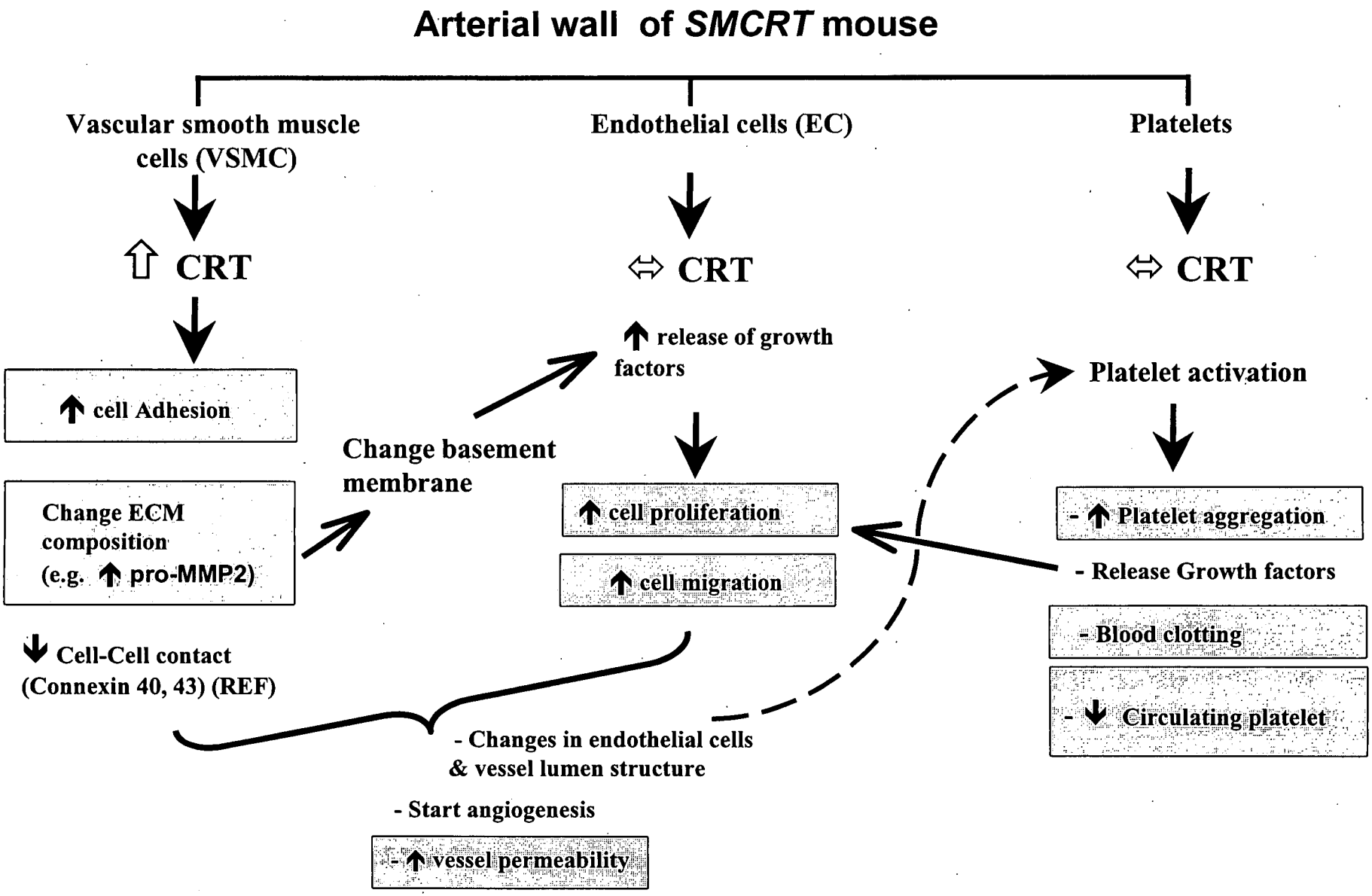
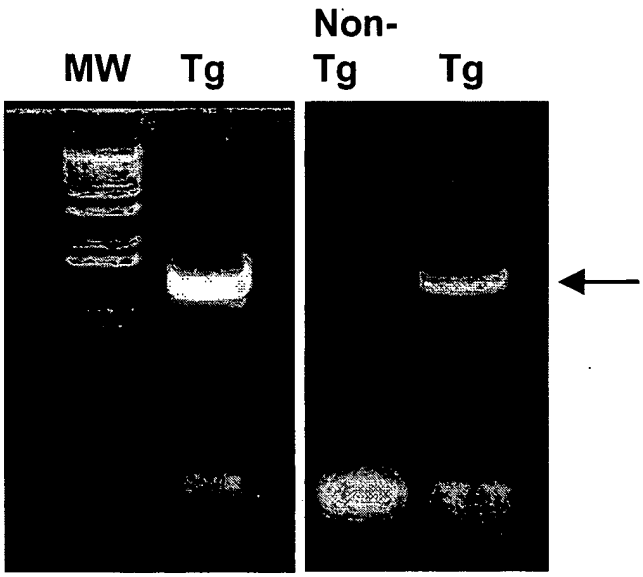

Arterial wall of SMCRT mouse
Vascular smooth muscle cells (VSMC)
Endothelial cells (EC)
Platelets
⇧ CRT
⇔ CRT
⇔ CRT
↑ release of growth factors
↑ cell Adhesion
Change basement membrane
Platelet activation
Change ECM composition (e.g. ↑ pro-MMP2)
↑ cell proliferation
- ↑ Platelet aggregation
↑ cell migration
- Release Growth factors
- Blood clotting
↓ Cell-Cell contact (Connexin 40, 43) (REF)
- ↓ Circulating platelet
- Changes in endothelial cells & vessel lumen structure
- Start angiogenesis
- ↑ vessel permeability

Fig. 1.

A



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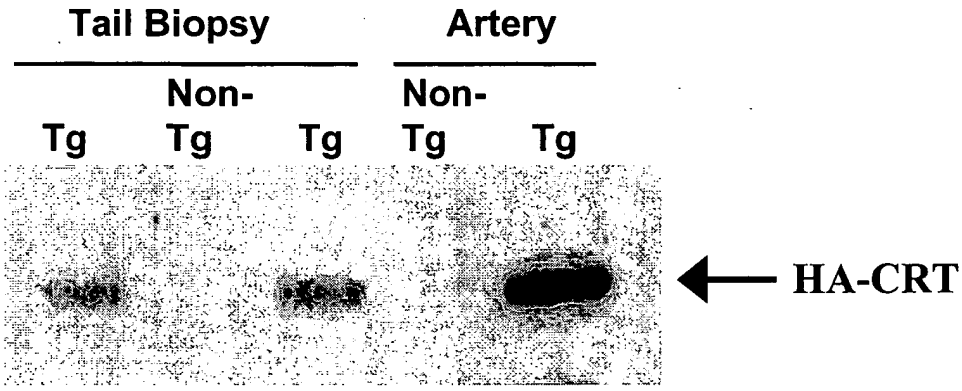
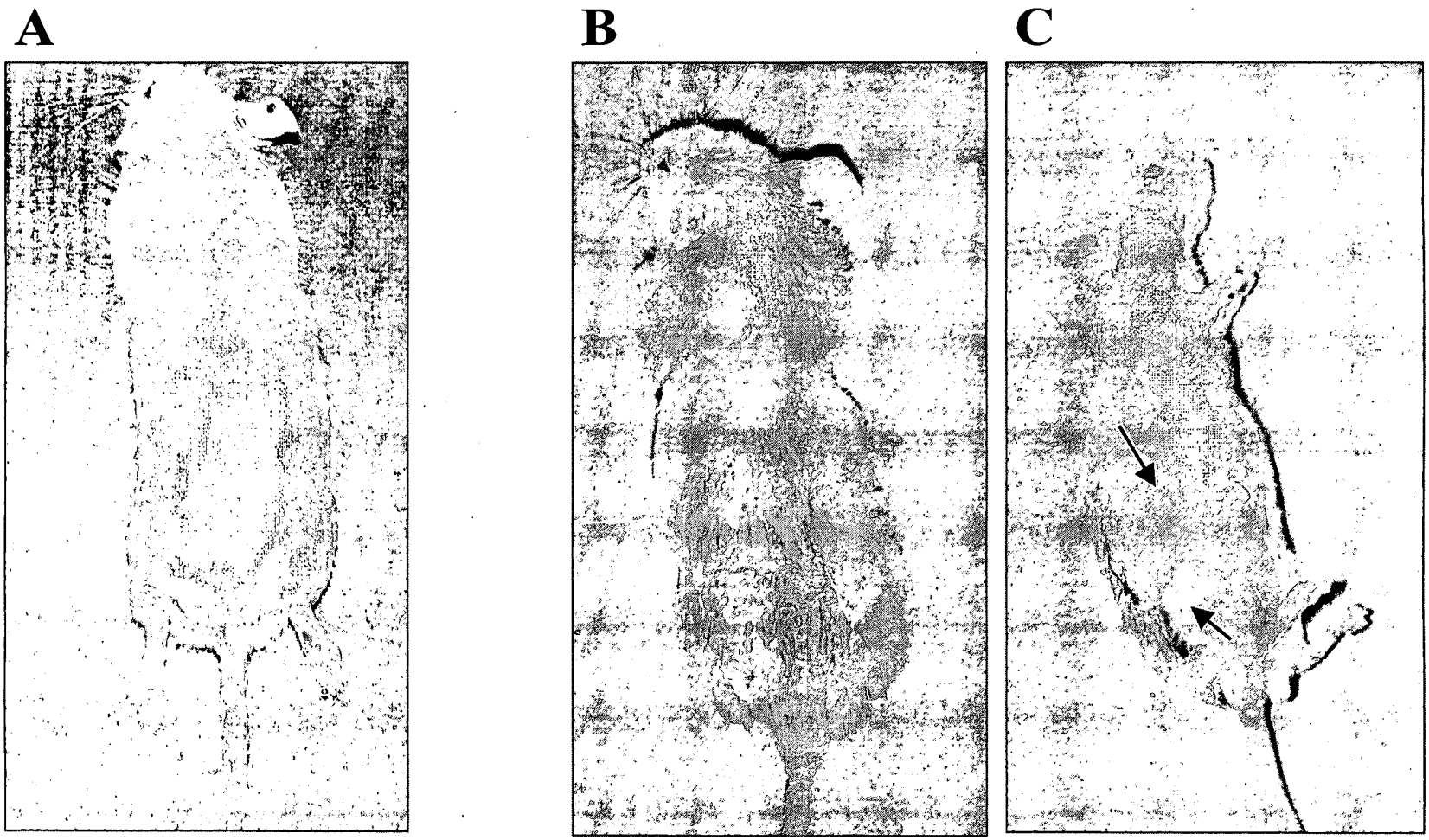


Fig. 2.



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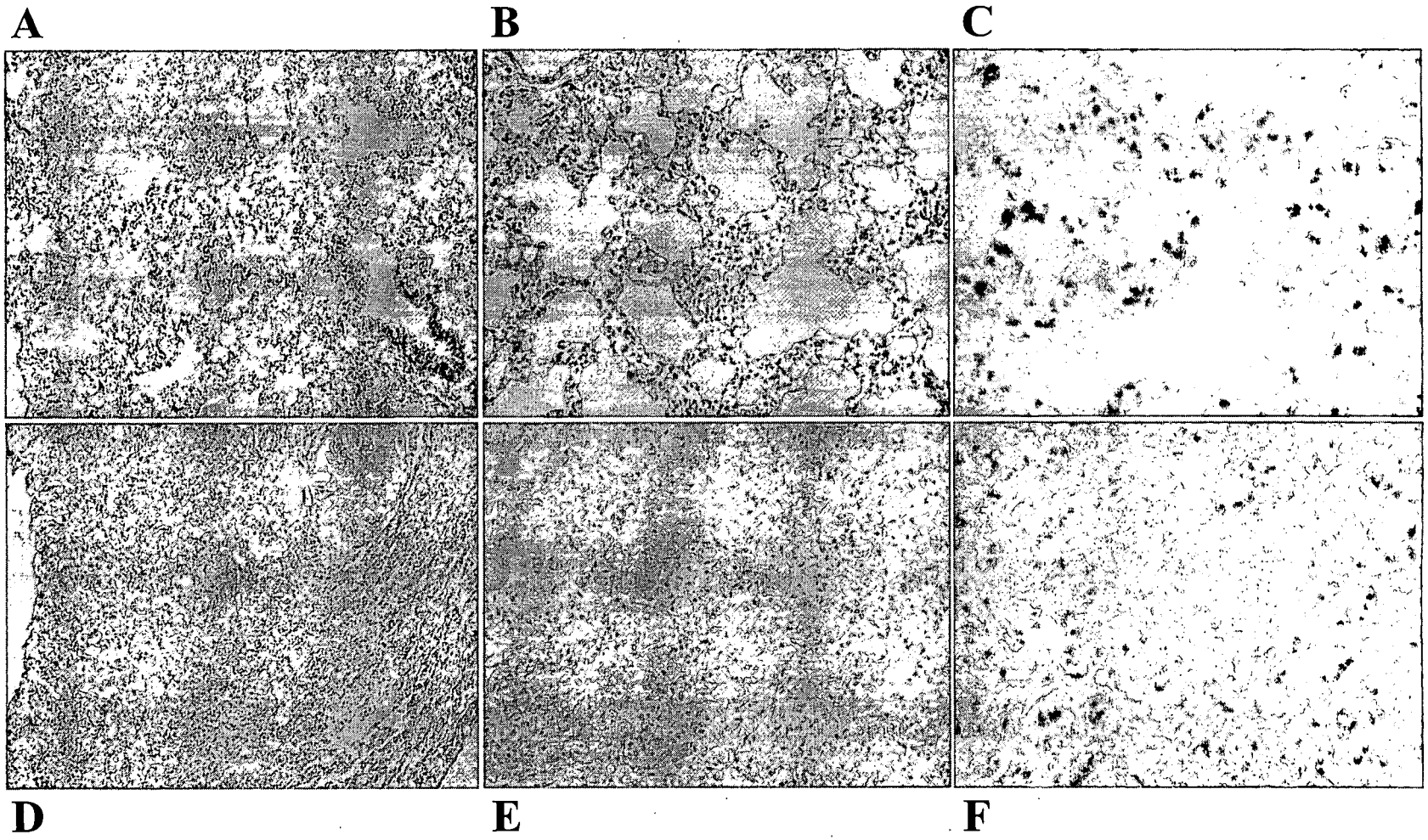


Fig. 4.

A. *SMCRT* Kidney

B. Wild type

C. *SMCRT*

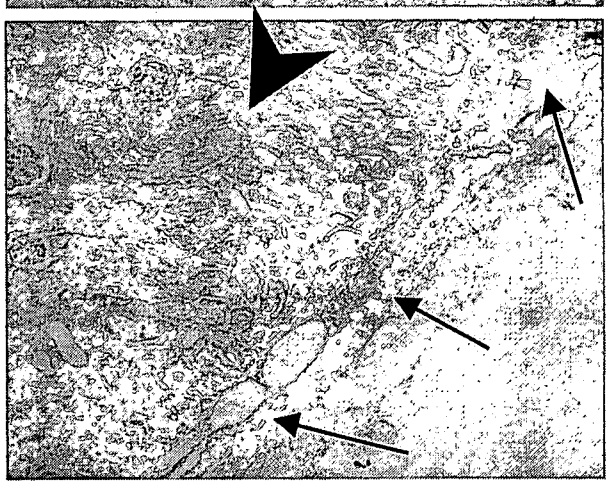


Fig. 5.

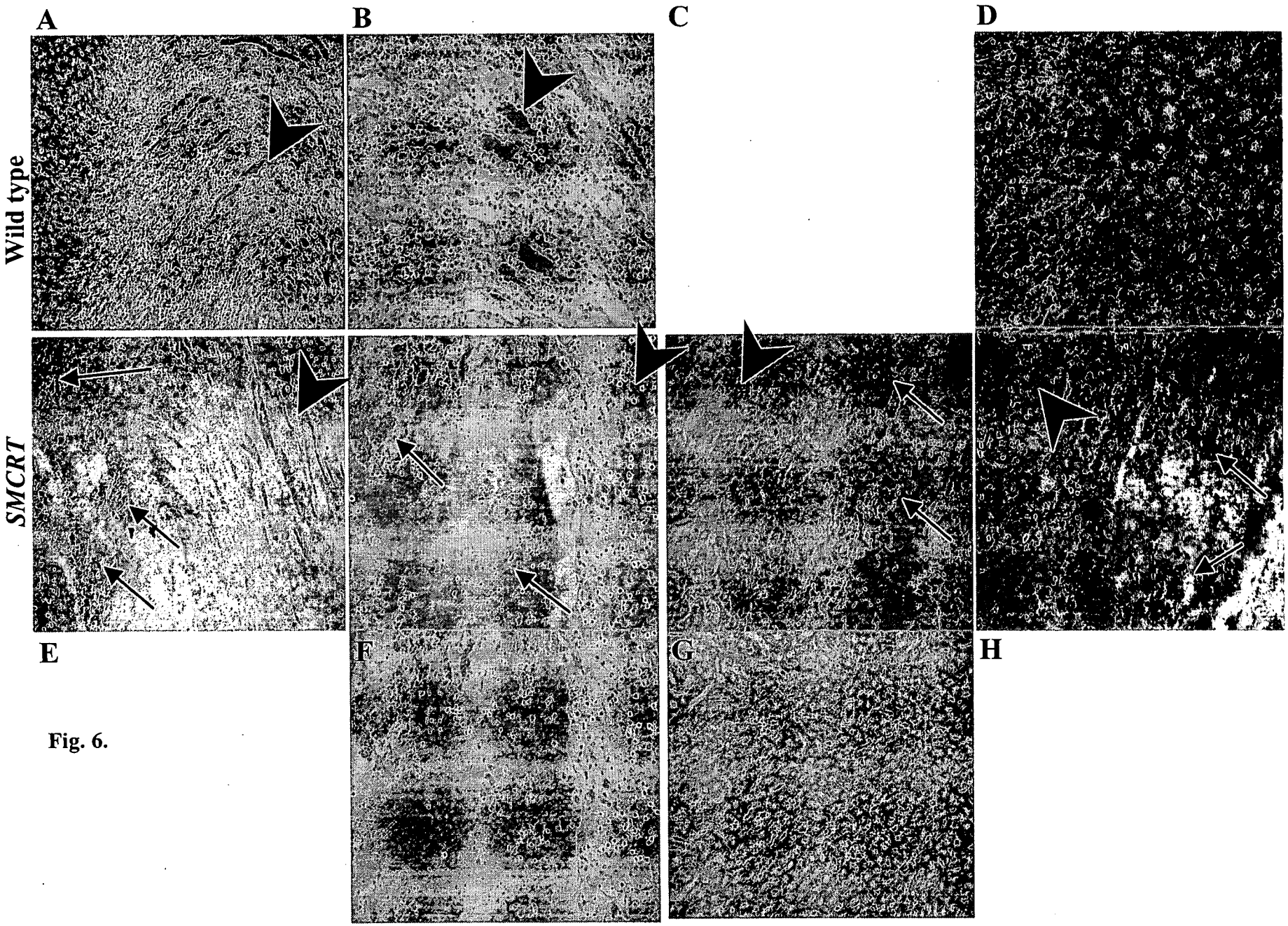


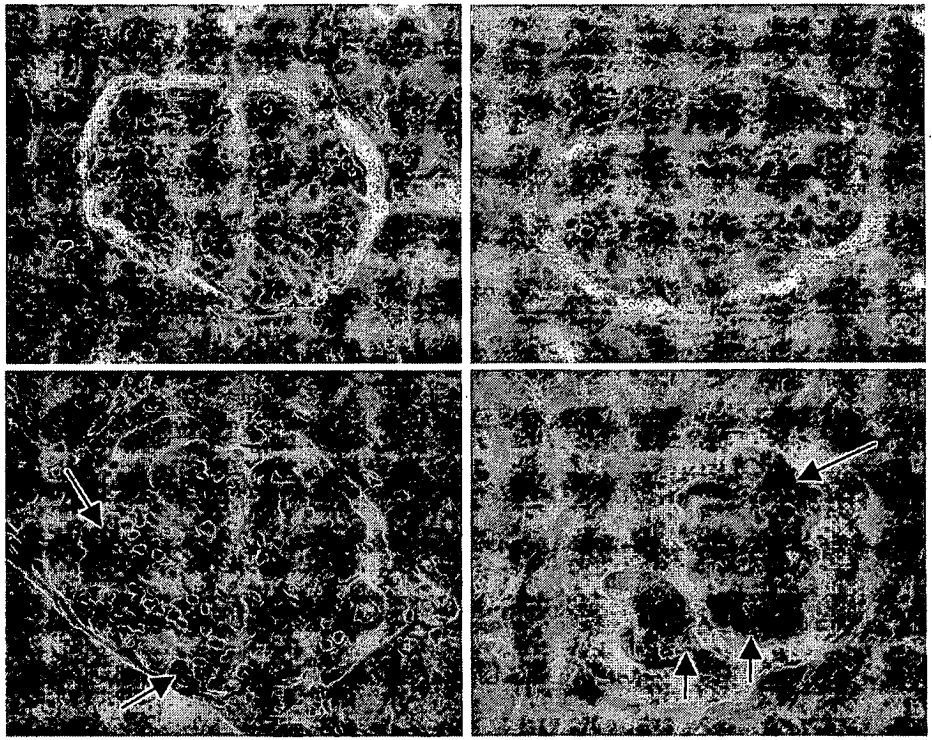
Fig. 6.

A.

Wild Type

B.

SMCRT



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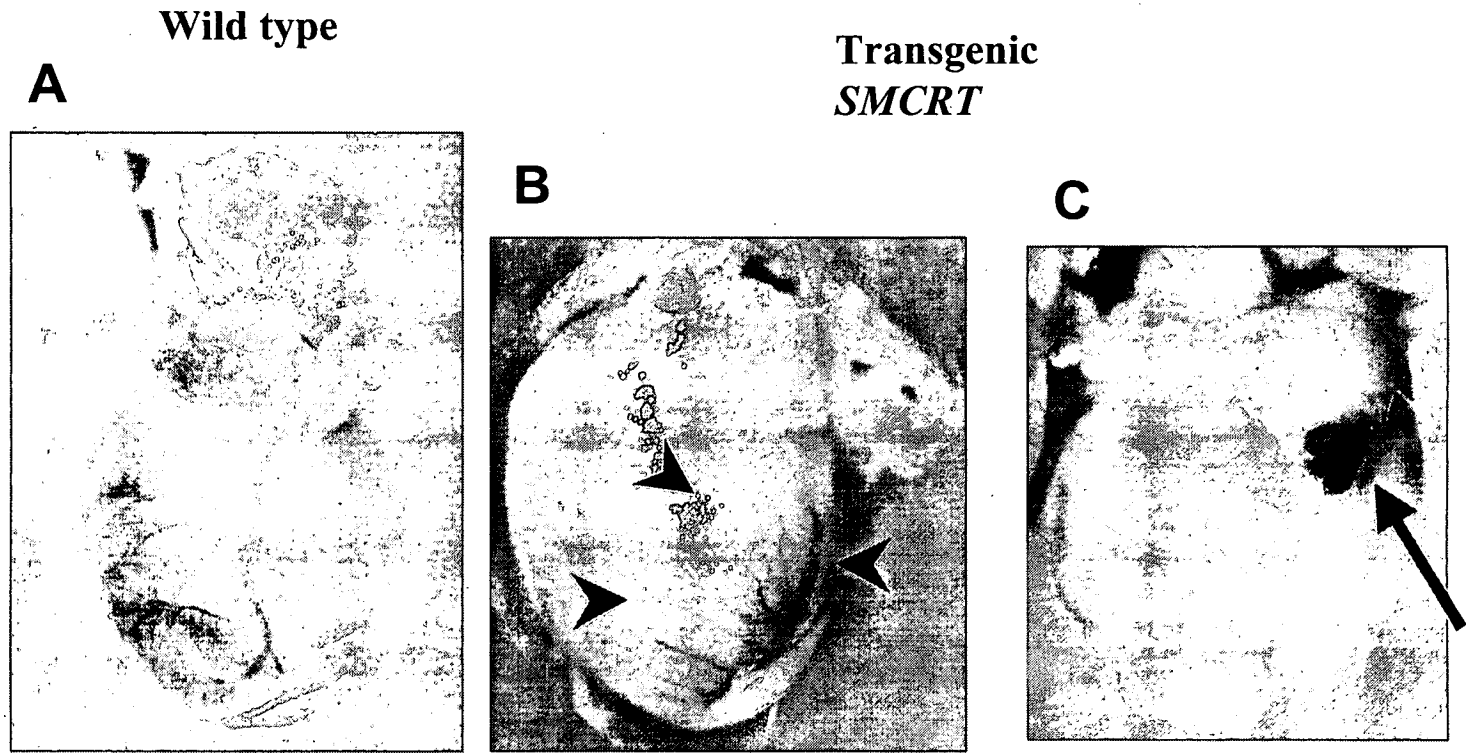


Fig. 8.

Wild type

SMCRT

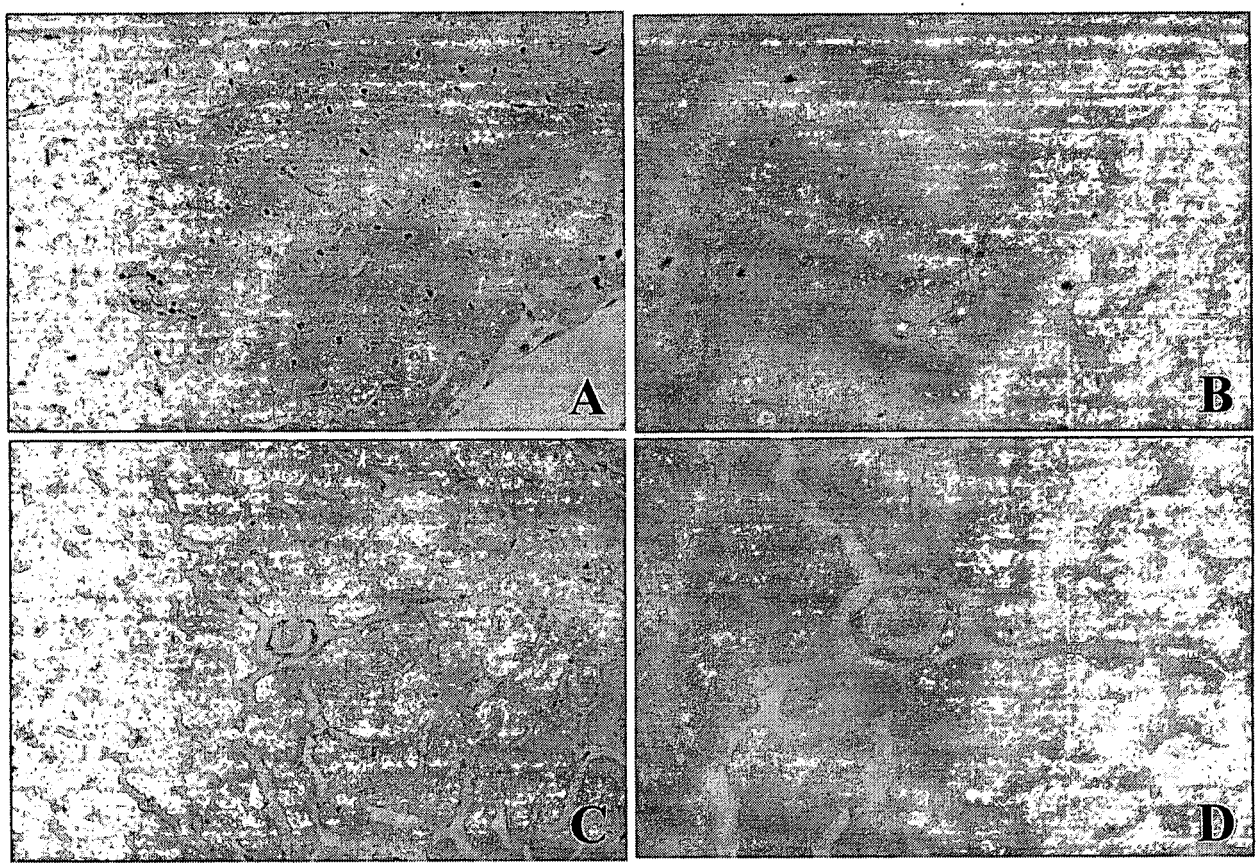


Fig. 9.

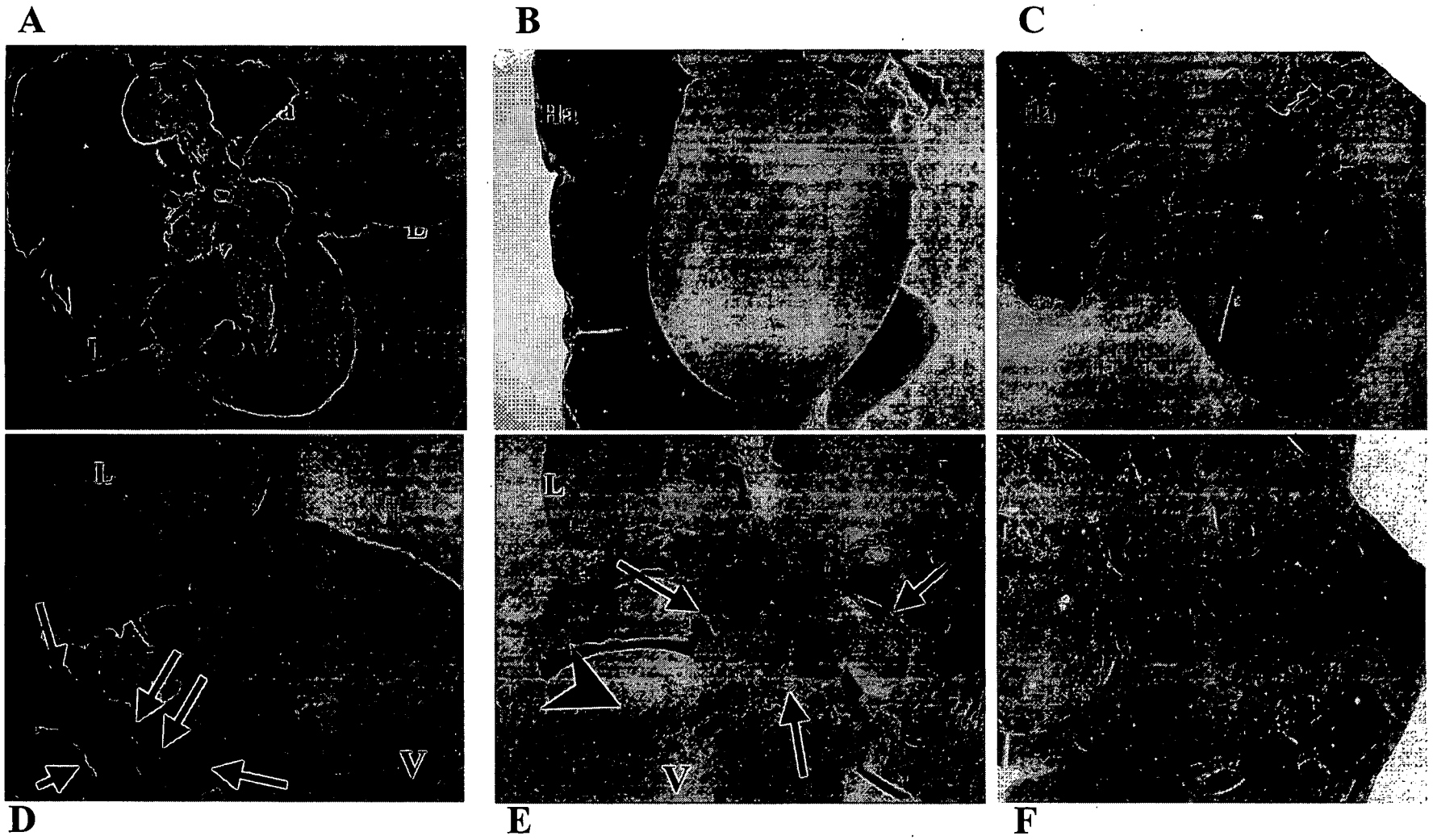
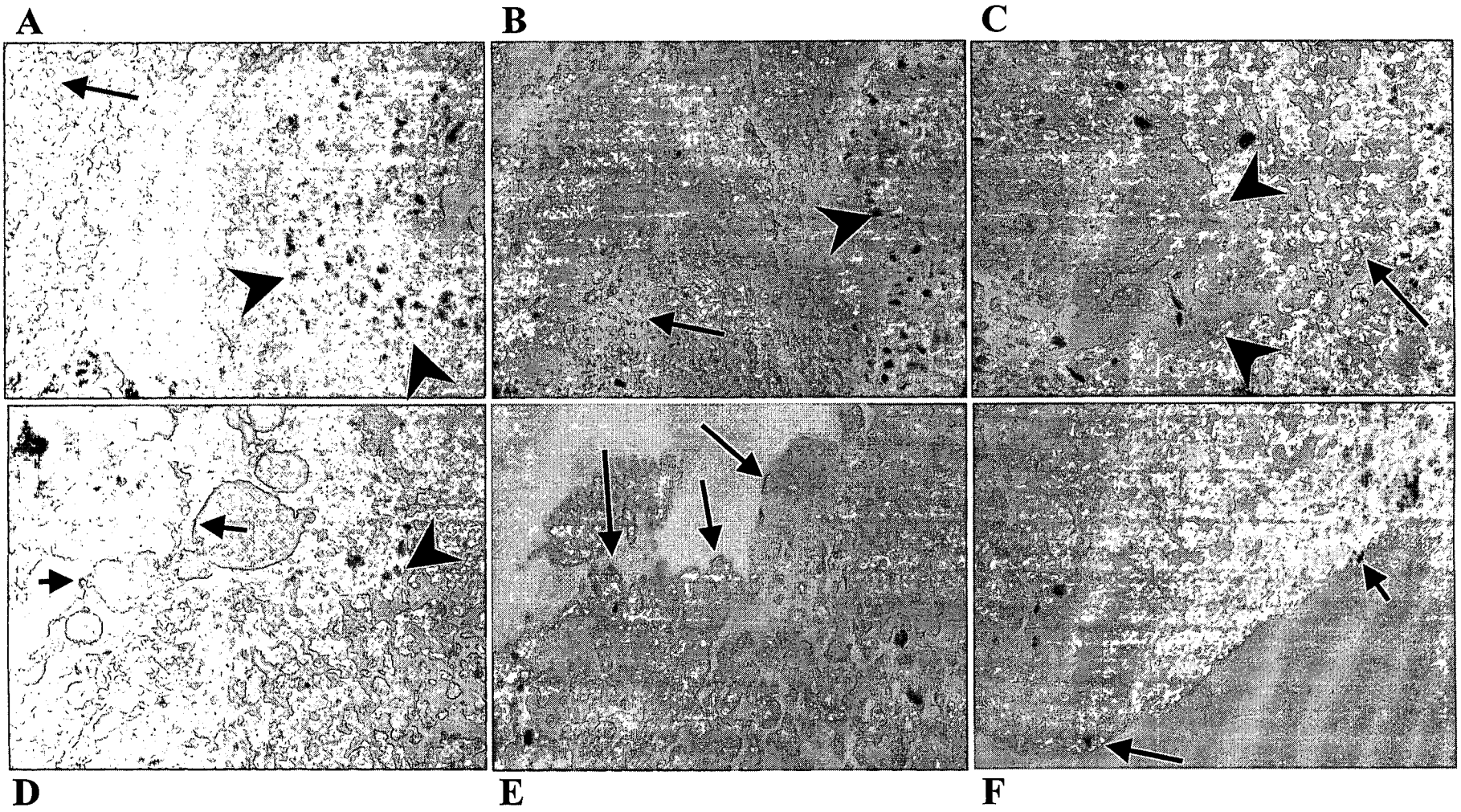


Fig. 10.



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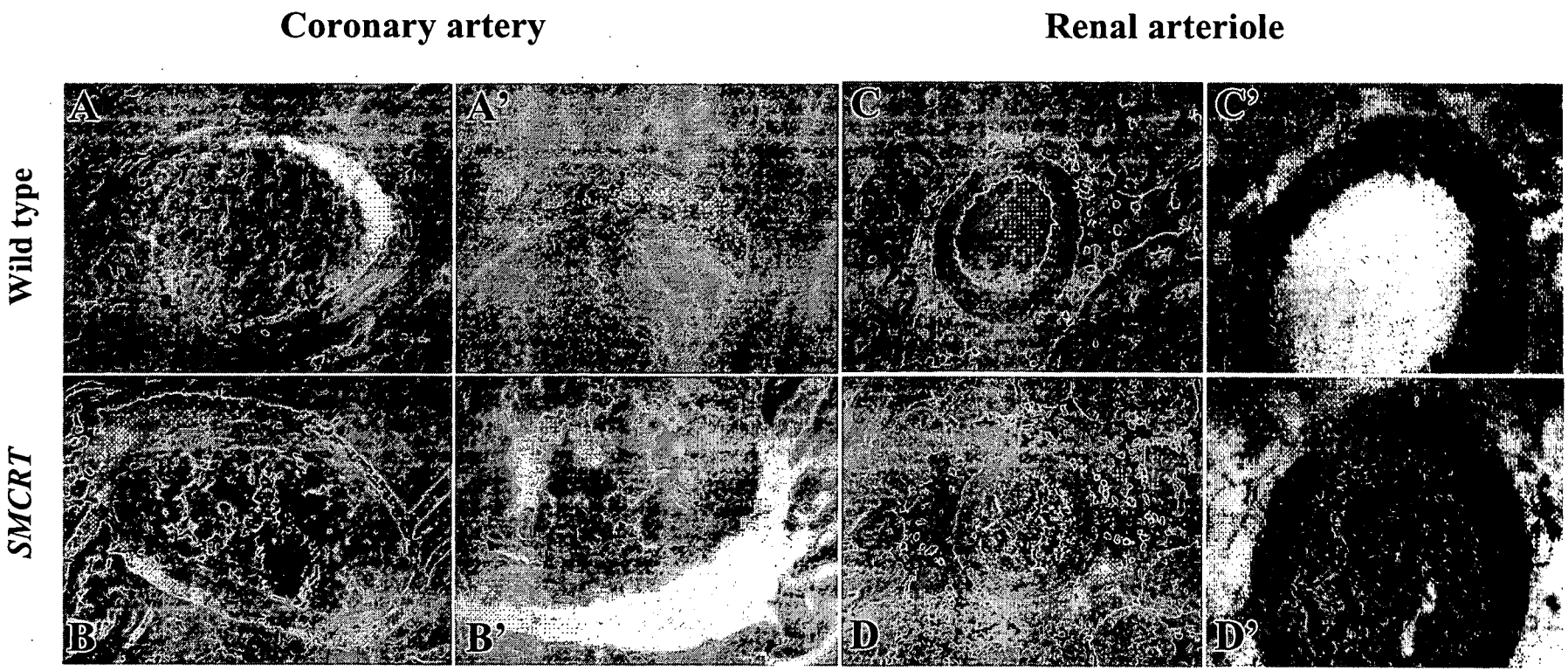

Fig. 12.

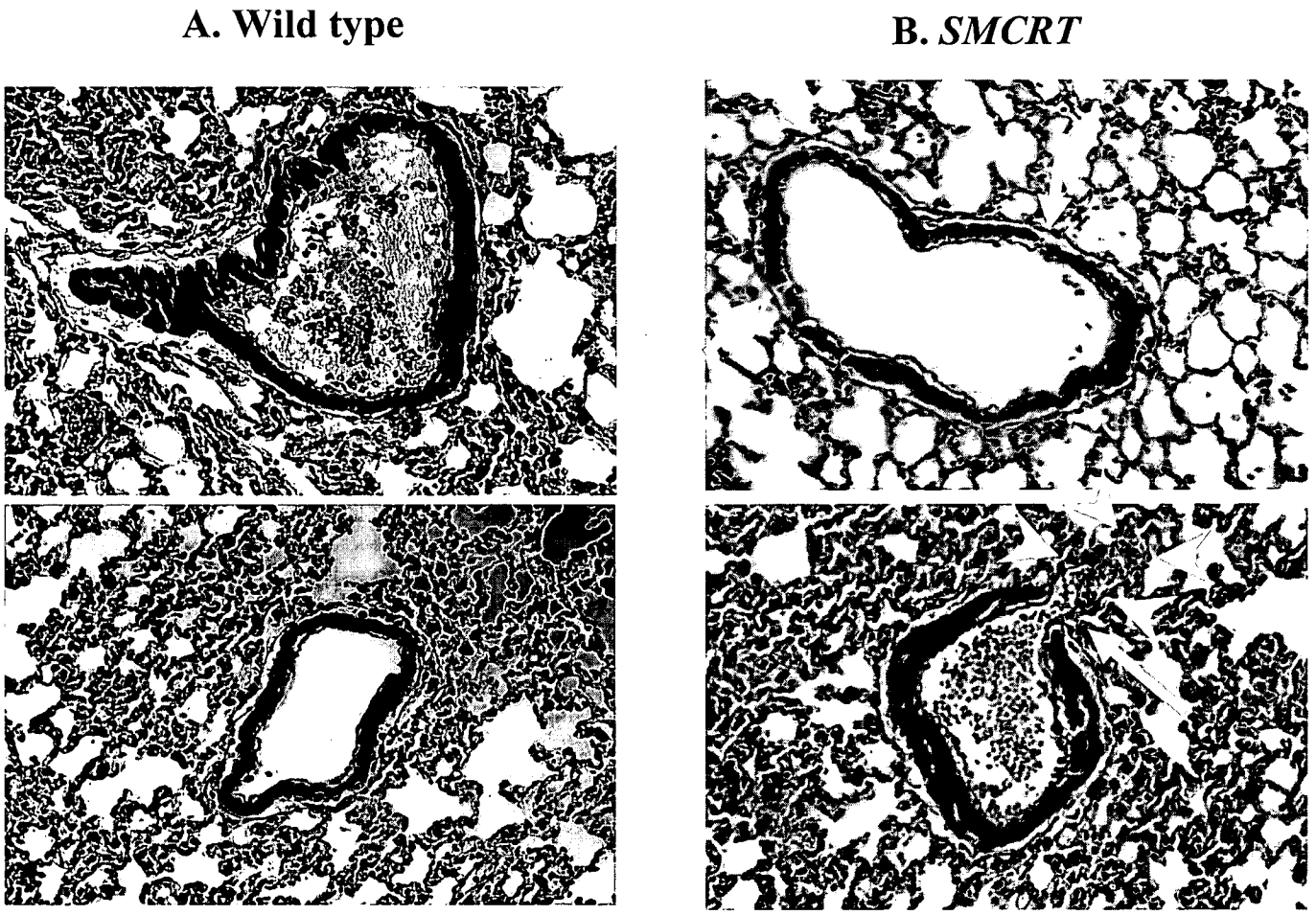

Figure 13- Masson Trichrome staining of lung sections of wild type (A) and *SMCRT* (B), showing the changes in the arteriole walls of the transgenic mice. Arrows (Green) indicates the disruption in the smooth muscle layer and migration of the endothelial cells thus developing a pouch containing red blood cells (lined by the arrow heads in B) which can lead to the formation of hemangioma.

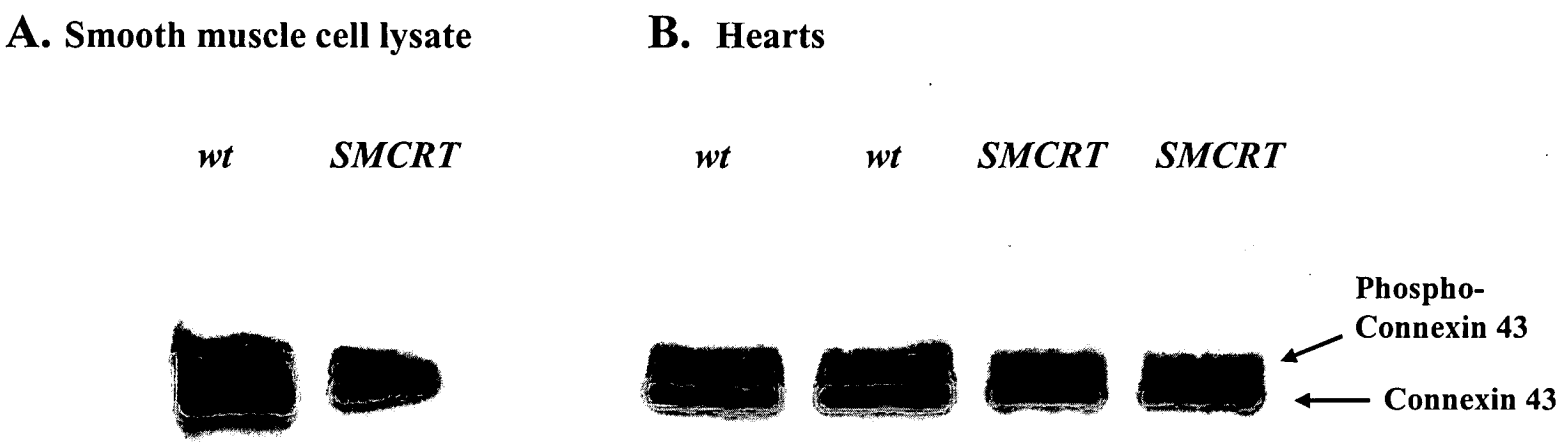

Figure 14- Western blot analysis showing connexion 43 expression in the vascular smooth muscle cells (A) and hearts (B) isolated from the wild type and *SMCRT* mice. There was a significant decrease in the connexion 43 protein in the transgenic mice as compare to the wild type mice.

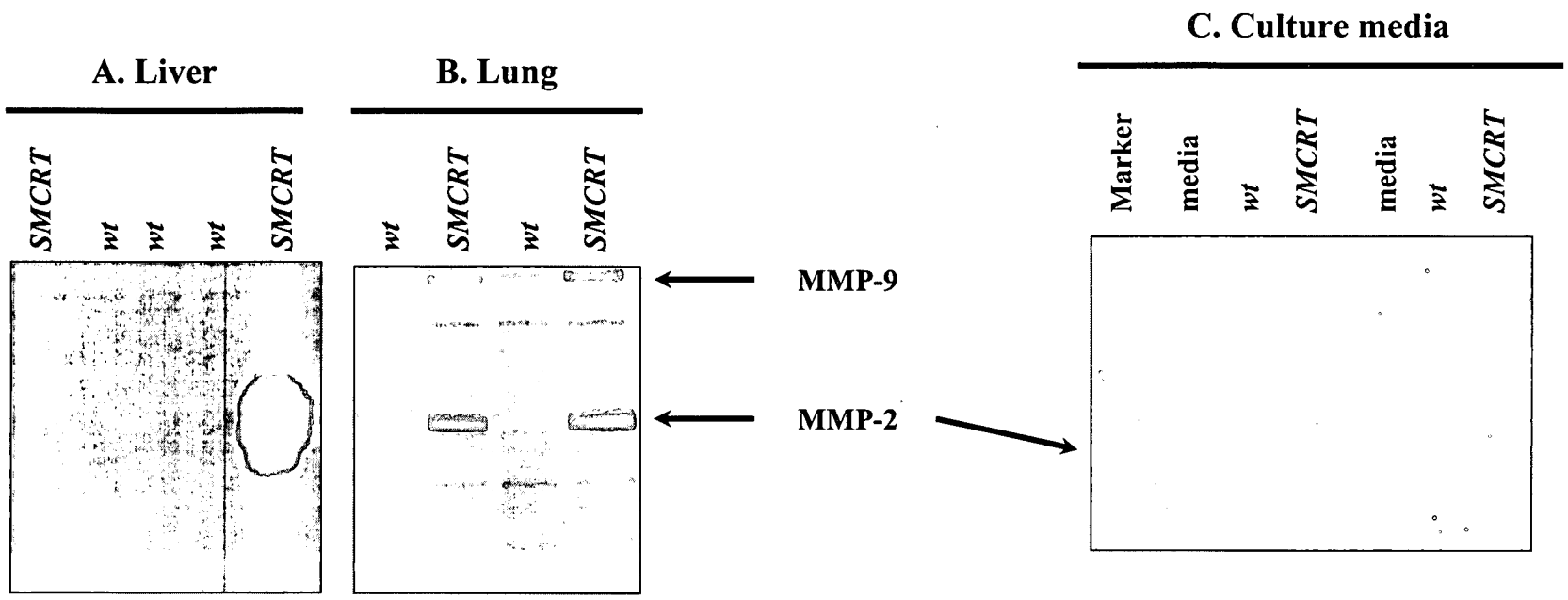

Fig. 15. Gelatin Zymography detecting the MMP-2 and MMP-9 activity in liver (A) and lung (B) tissue isolated from the *wt* and *SMCRT* mice. C) shows the activity of MMP-2 in the culture media from the *wt* and *SMCRT* smooth muscle cells detected by gelatin zymography. Briefly, cells were cultured in DMEM containing Insulin and transferrin for 24 hrs. 30 µl of media from culture plate with no cells (Media), wt cells and *SMCRT* cells (or 30 µg protein from each tissue) were separated on 7.5% SDS-acrylamide gel containing 1mg/ml Gelatin. After removal of SDS from the gel (to re-nature the proteins) , it was incubated in zymography buffer overnight at 37°C. The gels were then stained with Coomassie Blue and de-stained. The white bands represent the activity of MMP in the sample.

Figure 16
Nucleotide and protein sequence of SM22α–CRT

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Figure 17
Nucleotide and protein sequence of SM22α–CRT-HA

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